BIOLOGY AND IMMATURE STAGES OF PELINA TRUNCATULA, A CONSUMER OF BLUE-GREEN ALGAE (DIPTERA: EPHYDRIDAE)¹

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Abstract.—Information is presented on the life cycle and feeding habits of *Pelina truncatula* Loew, a common and widely distributed shore fly whose larvae feed on colonies of the blue-green algal genus *Cylindrospermum*. The egg, mature larva, and puparium are described and illustrated. A key is given that distinguishes the immature stages of *P. truncatula* from those of *Lytogaster excavata* (Sturtevant and Wheeler), another common inhabitant of *Cylindrospermum* colonies.

Containing at least 1200 species, the family Ephydridae is one of the largest entities within the acalyptrate Diptera. It is generally placed close to the family Drosophilidae in the superfamily Drosophiloidea (Griffiths, 1972). Although larvae of both families are basically microphagous in feed-ing habits, the drosophilids mostly utilize yeasts and other heterotrophic microorganisms, whereas the ephydrids primarily ingest autotrophic algal cells. Foote (1979) discussed the utilization of algae by different groups of Ephydridae and emphasized that certain species are trophically generalized (e.g. *Scatella stagnalis* (Fallén); Zack and Foote, 1978), while others are quite specialized (e.g. *Parydra quadrituberculata* Loew; Deonier and Regensburg, 1978).

This is the third contribution in a series dealing with the utilization of blue-green algae by ephydrid larvae. The first paper (Foote, 1977) presented general observations on the feeding habits of 12 species, and the second elucidated the life cycle and natural history of *Lytogaster excavata* (Sturtevant and Wheeler), a consumer of soil-inhabiting blue-green algae belonging to the genus *Cylindrospermum* (Foote, 1981). The present paper is concerned with another grazer of *Cylindrospermum*, *Pelina truncatula* Loew, a common and widely distributed species in shoreline and wetland habitats. Information is given on the life cycle and larval feeding habits, along with descriptions and illustrations of the egg, mature larva, and puparium.

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MATERIALS AND METHODS

Most of the field and all of the laboratory work was carried out in northeastern Ohio. Supplementary field observations were obtained in south central Arizona near Tucson and in northwestern Montana along the shores of Flathead Lake.

The laboratory rearings involved monocultures of 21 different species of algae that were maintained in an environmental chamber programmed to give a photoperiod of 15L:9D and a temperature of $22^{\circ}C$ ($\pm 1.0^{\circ}C$). Each algal culture was established on a nutrient agar substrate in sterile petri dishes, and larval feeding tests were performed utilizing procedures given by Zack and Foote (1978). Most of the algal monocultures were obtained from the University of Texas Culture Collection of Algae (Starr, 1978). Monocultures utilized in the larval feeding tests are listed below, along with their UTEX strain numbers. Algae lacking strain numbers were obtained from the Phycology Laboratory at Kent State University.

Cyanophyceae

Anabaena flos-aquae (1444) Anabaena variabilis (B377) Anabaena sp. Cylindrospermum sp. (LB942) Gloeocapsa alpicola (B589) Gloeocapsa sp. Lyngbya spiralis (B1831) Nostoc commune (584) Oscillatoria tenuis (B428) Oscillatoria sp. Phormidium sp. (1540) Synechococcus leopoliensis (625)

Chlorophyceae

Chlamydomonas eugametos (9) Chlamydomonas sp. Chlorella vulgaris (29) Cosmarium botrytis (175) Scenedesmus quadricauda (76) Vancheria sp.

Xanthophyceae

Botrydiopsis alpina (295) Botrydium becherianum (158)

Bacillariophyceae Navicula pelliculosa (668)

LIFE HISTORY

Although the genus *Pelina* contains seven species in the Nearctic Region (Clausen, 1973), none is presently recorded from the Neotropics (Wirth, 1968). It also occurs in the Palaearctic Region, with some six species listed for that area (Becker, 1926), and the Afrotropical Region (two species; Cogan, 1980). According to Clausen (1973), *P. truncatula* is the most common and widespread of the Nearctic species, having been recorded from throughout North America south of southern Canada. Although Clausen (1973) recognizes four subspecies, *P. t. truncatula* is the only one recorded from the eastern states, and most of the life history observations given here undoubtedly refer to that taxon. All descriptions of the immature stages are based on material collected in northeastern Ohio.

Deonier (1965) reported adults of *P. truncatula* to be occasional in marshreeds, sedge-meadow, and limnic-wrack habitats in Iowa, while Scheiring and Foote (1973) stated that the species was collected commonly in limnicwrack and mud-shore habitats in Ohio. I swept adults from the sandy margins of a drying tributary in Sabino Canyon near Tucson and from the sewage-laden shores of a small stream (Sonoita Creek) at Patagonia, Arizona. In northwestern Montana, adults were taken commonly from sandy shorelines of Flathead Lake and from low marshy areas bordering streams. Adults for rearing purposes were obtained in northeastern Ohio by sweeping over the shaded margins of the outlet of a package sewage plant. Soil algae, particularly species of blue-green genera, were abundant on the moist substrates in all collecting sites.

Adults were present throughout the warm season in the Ohio and Montana study sites and throughout the year in south central Arizona. The earliest record for adults in northeastern Ohio was obtained on April 25; the latest, on October 5.

A few data were obtained concerning adult longevity. One male and two females that were collected in nature near Kent, Ohio, on April 29 died on May 19, giving a longevity of at least 21 days. In contrast, laboratory reared adults rarely survived more than 15 days when confined in petri dishes containing monocultures of *Anabaena* sp. or *Cylindrospermum* sp. The premating period in a single reared pair was four days; the preoviposition period, six days. No courtship behavior was observed, and mating seemed to be of the assault type (Spieth, 1974).

Fecundity records were obtained from the two females collected in nature on April 29. One female deposited 28 eggs; the other, 37 over a 16-day period. No eggs were deposited by either female during the last five days of adult life. The potential fecundity probably is at least twice that obtained in the laboratory rearings, as a female collected in nature during early May possessed a total of 43 ovarioles, with each ovariole containing one nearly mature egg and 3-5 less developed oocytes. Assuming that each ovariole releases at least four eggs during the adult life of a female, the potential fecundity would exceed 170 eggs (4 × 43 ovarioles = 172 eggs).

In the laboratory, eggs were scattered widely into the blue-green algal cultures. In contrast to the white eggs of another blue-green feeding species, Lytogaster excavata (Foote, 1981), the eggs of P. truncatula possessed a distinct peach color. Also contrasting with L. excavata was the fact that the eggs of Pelina usually were placed below the alga-agar surface and frequently were out of contact with ambient air. Another difference was the papillose nature of the non-striated chorion (Fig. 13). The incubation period was 3-4 days (n = 18). Interestingly, newly hatched larvae tended to remain buried in the agar just below the algal mat rather than crawling over the algal surface. This tendency persisted throughout larval life, although older larvae occasionally were seen moving across the surface of algae. Supporting the laboratory observations was the fact that larvae were rarely seen on the surface of field-collected colonies of the blue-green algal genus Cylindrospermum. Instead, they were buried in the algal growth with only the posterior spiracles in contact with the ambient air. The habit of feeding below the surface of the algal growth is in distinct contrast to the behavior of species of Hvadina and Lytogaster which feed on the surface (Foote, 1977).

The results of larval feeding tests utilizing unialgal cultures are summarized in Table 1. Larvae completed development only on species of the bluegreen genera Anabaena, Cylindrospermum, Lyngbya, and Oscillatoria. Interestingly, not all genera of Cyanophyceae supported larval growth, and development was nil or greatly reduced in cultures of Gloeocapsa, Nostoc, Phormidium, and Synechococcus. Even among the blue-green species that allowed for larval development, there were apparent differences in nutritional suitability. For example, within the genus Anabaena high success rates were obtained on cultures of A. variabilis and an undetermined species (100% and 80%, respectively), whereas only 20% of the larvae reached the adult stage on A. flos-aquae. A strong indication of the trophic restriction of P. truncatula to blue-green algae is that no adults were obtained from larvae placed in cultures of any other group of algae. In fact, attempted culture on most of the non-cyanophyte algal species resulted in death of the larvae as first instars. Only cultures of Chlamydomonas sp., Cosmarium botrytis, Botrydiopsis alpina, Botrydium becherianum, and Navicula pelliculosa permitted some larval development. Of these five species, only C. sp., B. alpina, and B. becherianum supported growth to the third instar, and no larvae formed pupae.

Table 1. Results of larval feeding tests for *Pelina truncatula* using different algal monocultures.

		Percent Reaching Different Life Stages				
Alga		21.	3L	Р	А	
Cyanophyceae						
Anabaena flos-aquae	30	90	83	43	20	
Anabaena variabilis	20	100	100	100	100	
Anabaena sp.	10	100	80	80	80	
Cylindrospermum sp.	10	100	100	100	100	
Gloeocapsa alpicola	10	10	0	0	0	
Gloeocapsa sp.	10	100	90	0	0	
Lyngbya spiralis	5	100	40	40	40	
Nostoc commune	10	0	0	0	0	
Oscillatoria tenuis	20	30	30	30	30	
Oscillatoria sp.	10	50	50	40	10	
Phormidium sp.	20	0	0	0	0	
Synechococcus leopoliensis	10	30	0	0	0	
Chlorophyceae						
Chlamydomonas eugametos	10	30	0	0	0	
Chlamydomonas sp.	10	100	60	0	0	
Chlorella vulgaris	10	0	0	0	0	
Cosmarium botrytis	10	10	0	0	0	
Scenedesmus quadricauda	10	0	0	0	0	
Vaucheria sp.	10	0	0	0	0	
Xanthophyceae						
Botrydiopsis alpina	10	40	40	0	0	
Botrydium becherianum	10	60	10	0	0	
Bacillariophyceae						
Navicula pelliculosa	10	80	0	0	0	

As Table 2 indicates, the time required to complete larval and pupal development varied among the blue-green algal species that were nutritionally suitable. Thus, the combined larval-pupal period among larvae that fed on species of *Anabaena* and *Cylindrospermum* ranged between 16 and 21 days, whereas this period was extended to nearly 30 days in cultures of *Oscillatoria* sp. Nearly all of the slowed development occurred in the larval period which doubled from an average of 11.2 days to 21.5 days. Growth was fastest in *Anabaena* sp. which gave a combined larval-pupal period of ca. 16 days.

Similar results were obtained in nature. No immature stages were found in colonies of any alga except those of *Cylindrospermum* spp. Surprisingly, no larvae were taken in growths of certain of the blue-green genera that supported larval development in the laboratory rearings, suggesting that the genus *Cylindrospermum* is the primary, if not the sole, host in nature. Lar-

Alga	Larval Period			Pupal Period			Combined Period		
	n	Ĵ.	8	n	ĩ	S	n	Ĩ.	8
Anabaena flos-aquae	13	12.23	2.17	6	8.17	0.75	6	18.67	0.82
Anabaena variabilis	20	10.95	1.00	20	7.85	0.49	20	18.80	0.89
Anabaena Sp.	8	8.63	0.92	8	7.25	0.46	8	15.88	0.99
Cylindrospermum sp.	10	12.90	1.10	10	7.70	0.48	10	20.60	0.97
Lyngbya sp.	2	13.00	0.00	2	7.00	0.00	2	20.00	0.00
Oscillatoria tenuis	6	10.67	0.52	6	7.33	0.52	6	18.00	0.89
Oscillatoria sp.	4	21.50	1.00	1	8.00		1	29.00	

Table 2. Developmental times in days for *Pelina truncatula* feeding on different algal monocultures.

vae of two other hyadinine genera, Hyadina and Lytogaster, were also found in colonies of Cylindrospermum. However, only rarely did larvae of Pelina co-occur with species of these other genera in the same algal colony. In general, Pelina larvae were taken from colonies growing in somewhat wetter habitats, whereas larvae of Hyadina and Lytogaster were usually associated with soil-inhabiting patches of Cylindrospermum. Thus, at least ten larvae of P. truncatula but none of the other two genera of Hyadinini were discovered in a floating mat of mixed algae occurring in a shallow pool of a small stream in southern Arizona. Further, only Pelina larvae were found in a submerged colony of *Cylindrospermum* in a stand of cattail (*Typha latifolia* L.) growing along the shores of Flathead Lake in Montana. However, segregation into different habitats was not absolute, as Pelina larvae were occasionally discovered in soil-inhabiting Cylindrospermum. Thus, four larvae of L. excavata and five of P. truncatula were found in two petri dish samples (2.84 cm²/dish) of *Cylindrospermum* that was growing on moist. sewage-laden soil bordering the drainage ditch of a small package sewage treatment plant near Kent, Ohio, on June 3, 1972.

Larvae moved about slowly when feeding on a suitable blue-green algal colony (e.g. *Cylindrospermum*) but became more active and soon abandoned a nutritionally unfavorable alga (e.g. *Chlorella*). During feeding the larvae moved the anterior end of the body back and forth as they crawled across or through the algal substrate. Trichomes of blue-green algae were ingested whole or were torn away from the algal matrix by the ripping action of the mouthhooks working in conjunction with the comblike structures located around the oral opening (Fig. 5). These structures raked across the algal surface, shredding the matrix, and pulling entire trichomes or fragments of trichomes into the oral cavity. The floor of the pharyngeal sclerite has nine low, relatively flat-topped ridges (Fig. 14), but it is doubtful if these structures can serve as filtering devices. In this respect, *Pelina* larvae resemble those of *L. excavata* (Foote, 1981). During feeding a steady stream

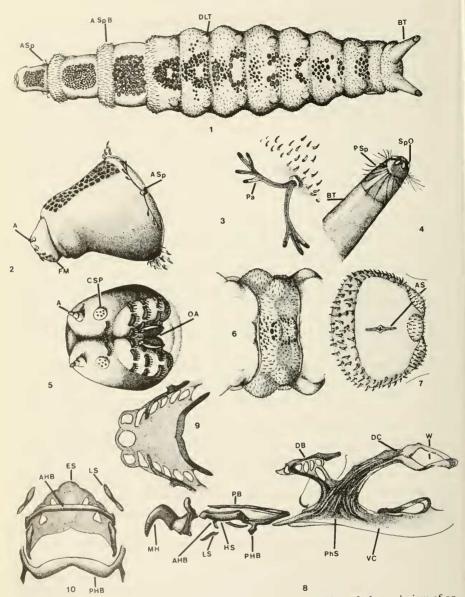
Flight period	Mid-May to late October
Adult longevity	21 + days
Premating period	4 days
Preoviposition period	6 days
Incubation period	3–4 days
Larval period	11–14 days
Pupal period	7–9 days
Length of life cycle	28–33 days
Fecundity	$170 \pm$
Number of generations/yr	5-6

Table 3. Life cycle data for *Pelina truncatula* in northeastern Ohio. Rearings maintained at 22°C, with *Cylindrospermum* sp. serving as adult and larval food.

of algal material entered the oral opening and progressed steadily down the alimentary canal. Although the mouth parts moved fairly rapidly, the "flickering" movement typical of such filter-feeding ephydrid larvae as *S. stagnalis* (Zack and Foote, 1978) was not noticeable. Larvae frequently were buried in the agar substrate except for the posterior spiracles which usually retained contact with the surface. Buried larvae fed by bending up the anterior third of the body until the facial mask came into contact with the algal mat.

Several larvae usually fed together within each algal colony, and no obvious aggressive behavior between individuals was noted. Five larvae were found together in a 7.5 cm² patch of *Cylindrospermum* growing on moist soil in northeastern Ohio, and over 10 mature larvae and 15 puparia were collected within a 144 cm² area of a floating algal mat in south central Arizona. The larval period lasted 11–14 days and averaged 12.9 days under laboratory conditions when *Cylindrospermum* sp. served as the larval food (n = 10). Pupation usually occurred within the algal colony, although a few puparia were found on the sides of the petri dishes above the algal surface. All field-collected puparia were taken from *Cylindrospermum* colonies. In all cases, the posterior spiracles were in contact with ambient air, even though most of the puparium was buried in the algal mass. The pupal period lasted 7–9 days and averaged 7.7 days (n = 10).

Assuming a preoviposition period of 6.0 days, an incubation period of 3.5 days, a larval period of 12.9 days, and a pupal period of 7.7 days, the life cycle can be completed in approximately 30 days. With a warm season lasting approximately 150 days in the latitude of northern Ohio (mid-May to mid-October), five generations a year could be produced in the northern states (Table 3). The discovery of larvae and puparia during November and early April in southern Arizona suggests a continuous cycling of generations in that area of the country.



Figs. 1–10. *Pelina truncatula*, third-instar larva. 1, Dorsal habitus. 2, Lateral view of anterior end. 3, Anterior spiracle. 4, Posterior spiracle. 5, Facial mask. 6, Dorsal view of segment 9, 7, Perianal pad. 8, Lateral view of cephalopharyngeal skeleton. 9, Dorsal bridge of pharyngeal sclerite. 10, Ventral view of hypostomal sclerite. Abbreviations: A = antenna; AHB = anterior hypostomal bridge; AS = anal slit; ASp = anterior spiracle; ASpB = anterior spinule band; BT = breathing tube; CSP = circular sensory plate; DB = dorsal bridge; DC = dorsal cornu; DLT = dorsolateral tubercle; ES = epistomal sclerite; FM = facial mask; HS = hy-

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DESCRIPTIONS OF IMMATURE STAGES

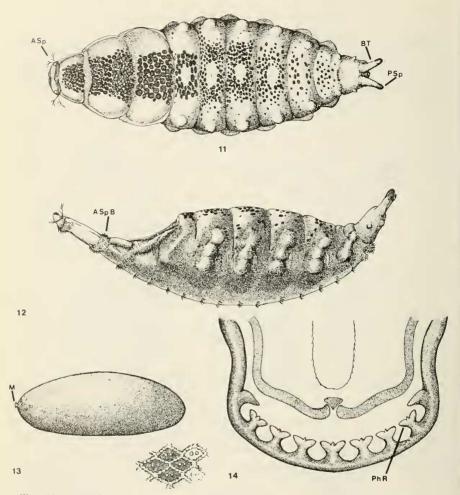
Egg (Fig. 13).—Length 0.45–0.52 mm, $\bar{x} = 0.50$; width 0.18–0.22 mm, $\bar{x} = 0.20$ (n = 10). Elongate-ovoid, slightly flattened ventrally. Pinkish in color when living, white when preserved. Chorion appearing papillose, with reticulated pattern; micropylar end not upturned, bearing small tubercle apically; opposite end rounded and not upturned.

Mature third-instar larva (Fig. 1).—Length 4.0–5.5 mm, $\bar{x} = 4.6$; width 0.8–1.1 mm, $\bar{x} = 1.0$ (n = 10). Somewhat flattened dorsoventrally; anterior end tapering, posterior end blunt and bearing apically 2 diverging breathing tubes; margins of body segments smooth, without noticeable lateral indentations; single row of low tubercles dorsolaterally on segments 5–11. Integument dorsally with conspicuous black scales; scales particularly noticeable on segments 2–5, less so on remaining segments; anterior borders of segments 3–5 with rows of dark bristles, bristle rows distinctly less conspicuous on remaining segments.

Segment 1 (pseudocephalic) (Fig. 5) frequently invaginated into segment 2, bearing antennae apically, circular sensory plates apicoventrally, and facial mask around oral aperture ventrally; antennae appearing 3-segmented; circular sensory plates with complete rims, each plate bearing 5-6 peglike structures; facial mask (Fig. 5) with conspicuous rows of comblike structures bordering oral aperture, 1 row anterior to aperture, others lateral to opening, each structure composed of narrow piece anteriorly and several narrow teeth posteriorly. Segment 2 (prothoracic) (Fig. 2) with numerous black scales dorsally, bearing bifurcate anterior spiracles near posterolateral border; each spiracle (Fig. 3) with 2 strongly diverging branches, dorsal branch bearing 4-5 elongate papillae apically, ventral branch with 3 apical papillae, base of spiracle arising from slightly pigmented ringlike structure. Segments 3-11 similar except in dorsal patterns formed by darkened tegumentary scales (Fig. 6); dorsolateral tubercles best developed on segments 5-11; venter of each segment with creeping welts bearing blackened bristles apically.

Segment 12 (caudal) bearing perianal pad ventrally and spiracular breathing tubes apicolaterally; perianal pad (Fig. 7) semicircular, bearing anal slit medially, pad bordered by several rows of spinules, post-anal spinule pad well-developed. Breathing tubes (Fig. 4) elongate, approximately $3 \times$ as long as wide, tapering distally; each tube capped by spiracular plate, plates with 4 radiating spiracular openings and 4 groups of branching hairlike processes.

postomal sclerite; LS = ligulate sclerite; MH = mouthhooks; OA = oral aperture; Pa = papilla; PB = parastomal bar; PhS = pharyngeal sclerite; PHB = posterior hypostomal bridge; PSp = posterior spiracle; SpO = spiracular opening; VC = ventral cornu; W = window.



Figs. 11–14. *Pelina truncatula*, immature stages. 11, Dorsal view of puparium. 12, Lateral view of puparium. 13, Egg. 14, Pharyngeal ridges of mature larva. Abbreviations: M = micro-pylar end; PhR = pharyngeal ridge; other abbreviations as in Figs. 1–10.

Cephalopharyngeal skeleton (Fig. 8) length 0.44–0.46 mm, $\bar{x} = 0.45$ (n = 10). Mouthhooks paired, not connected dorsally; hook part narrow and sicklelike, without accessory teeth: basal part broader, with small window ventrally and narrow projection posterodorsally. Hypostomal sclerite (Fig. 10) composed of lateral rods connected by 2 narrow, arched hypostomal bridges, anterior bridge evenly curved, posterior bridge indented medioposteriorly. Epistomal sclerite (Fig. 10) broad, with 4–6 small windows laterally. Parastomal bars (Fig. 8) narrow, seemingly not connected posteriorly to pharyngeal sclerite, extending anteriorly above hypostomal sclerite, anterior ends not expanded or connected. Pharyngeal sclerite (Fig. 8) largely pigmented; dorsal cornua rather broad, connected anteriorly by dorsal bridge, each cornu with large, irregular window posteriorly; dorsal bridge (Fig. 9) broad, with numerous windows laterally; ventral cornua broad, each with narrow window posterodorsally; floor of pharyngeal sclerite with 9 rather broad ridges (Fig. 14), lateral ridges incomplete, middle 7 ridges bearing lateral lamellae apically.

Puparium (Figs. 11, 12).—Length 4.0–4.6 mm, $\bar{x} = 4.2$; width 0.9–1.2 mm, $\bar{x} = 1.0$ (n = 10). Swollen dorsoventrally at midlength, anterior and posterior ends tapering and distinctly upturned; anterior end somewhat invaginated and bearing branched anterior spiracles apicolaterally; posterior end bearing 2 somewhat diverging breathing tubes apically. Dorsum of more anterior segments bearing numerous, densely clustered black scales, scales increasingly scattered on more posterior segments. Perianal pad somewhat invaginated; ventral creeping welts noticeable as bands of blackened bristles.

DISCUSSION

The discovery that at least nine species of four genera (*Axysta*, *Hyadina*, *Lytogaster*, *Pelina*) of the ephydrid tribe Hyadinini have larvae that feed on blue-green algae suggests strongly that this tribe is trophically unified by its preference for this group of microorganisms. In contrast, species of the three other North American genera currently assigned to the Hyadinini have distinctly different larval feeding habits. Thus, larvae of *Brachydeutera* appear to feed on decomposing plant remains in shallow pools (Williams, 1939), those of *Ochthera* are predacious (Simpson, 1975), and *Gastrops* larvae have been found in the eggs of frogs (Bokerman, 1957). Perhaps these three genera at least should be removed from the Hyadinini.

The immature stages of *L. excavata* and *P. truncatula*, two species commonly found in colonies of *Cylindrospermum*, are quite distinctive and can be separated by use of the key given below.

Key to Immature Stages of Lytogaster excavata and Pelina truncatula Egg

1. Ends of egg upturned, chorion striated; living embryo white

Ends of egg not upturned, chorion papillose; living embryo peach colored P. truncatula (Sturtevant and Wheeler)

Mature Larva, Puparium

1. Dorsum without conspicuous black scales; branches of anterior spiracles with sessile papillae L. excavata Loew Dorsum with conspicuous black scales; branches of anterior spiracles with papillae on elongate finger-like stalks

..... P. truncatula (Sturtevant and Wheeler)

The utilization of blue-green algae by ephydrid larvae may have considerable practical as well as ecological significance, as many species of this group of algae are capable of fixing nitrogen in aquatic and terrestrial habitats (Balandreau et al., 1975; Granhall, 1975; Maque, 1977). Because some ephydrid larvae feed exclusively on cyanophytes and are occasionally quite abundant, they may have an unsuspected impact on the amount of nitrogen fixed by blue-green algal colonies. The presence of numerous larvae of hyadinine species within colonies of soil-inhabiting blue-green algae can result in fairly rapid destruction of the algal growths. As many as 47 larvae of *L. excavata* have been found in one petri dish sample (2.84 cm²) of *Cylindrospermum* sp. growing on moist soil in southern Arizona. Larvae of Hyadina, *Lytogaster*, and *Pelina* readily abandon a *Cylindrospermum* colony once it has been largely consumed and move to adjacent colonies. Thus, their cumulative effect on soil-inhabiting blue-greens could be considerable.

At least four species of Ephydridae have larvae that deed on blue-green algae occurring as floating algal mats in shallow-water habitats (Foote, 1977). Larvae of *Setacera pacifica* (Cresson) occasionally became so abundant in floating colonies of *Anabaena* sp. in alkaline ponds in northwestern Montana that the mats quickly became riddled. As a result, the mats were easily disrupted and dispersed by wave action. It is of more than passing interest in this connection that the bacterium responsible for Legionnaire's disease has been reported to be associated with floating mats of such blue-green algae as *Fischerella* sp., *Phormidium* sp., and *Oscillatoria* sp. (Tison et al., 1980).

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